REVIEW ARTICLE

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Biosimilars for psoriasis: preclinical analytical assessment to determine similarity

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Summary

Biosimilars, sometimes called 'generic biologics', are no longer a vision for the future but a present-day reality. Drug manufacturers and regulatory authorities are charged with ensuring that these products are safe and effective. Because biologically produced medications are large, complex proteins, many factors affect the quality of the end product, including glycosylation and presence of impurities, and thus many factors need to be compared between an emerging biosimilar and its originator biologic. Indeed, preclinical analytical assessments to determine similarity to an originator biologic are critical and are considered to be the foundation for regulatory approval of biosimilars. Here, the science behind the preclinical development of biosimilars is discussed by members of the International Psoriasis Council, and suggestions are put forth to try to ensure that future biosimilars are produced in a high quality and standardized manner.

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What's known already about this topic?

- Biosimilars are biological drugs that are similar to an original marketed biological product.
- Approximately 40 different analytical methods are utilized to assess approximately 100 different drug attributes.
- Preclinical analytical assessments are used to determine similarity to an originator biologic and are critical for regulatory approval of biosimilars.

What does this study add?

- The International Psoriasis Council suggests guidelines for standardization of preclinical assessments of emerging biosimilars through the development of a biosimilar index.

Biosimilar drugs, or biosimilars, are biological drugs that are similar to an original marketed biological product.[1] They have also been described as 'generic biologics', although this term is technically inaccurate, as generic drugs are not just similar, but identical, to originally marketed products. Biologics are manufactured using unique bioengineered cell lines that produce the drugs. Companies in the business of making biosimilars are not in possession of the original cell line utilized for the originator compound, and thus their biologic, derived from a new cell line, is not identical to the original product. The basic biosimilar definition put forth here then urges the question of 'How is similarity defined?'[2] Unfortunately, the answer to this question is not simple and is not being answered in the same way by different biosimilar manufacturers and drug-regulating authorities.

Creation and preclinical analysis of a new biosimilar compound involves many steps.[3] At each step, the new biosimilar drug is compared and contrasted with the original. Instead of relying on any one key piece of data, the weight of all the analytical assessments is considered when determining whether the biosimilar is 'similar' to the originator compound.[4] In the current article, members of the International Psoriasis Council discuss (i) how biosimilars are created; (ii) the variety of analytical methods being used to determine how similarity is being defined prior to clinical studies; and (iii) suggested guidelines for standardizing these processes in the future. Defining efficacy and safety similarity between biosimilars and originator biologics through clinical studies, as well as examination of how biosimilars are affecting dermatology clinical practice, are important topics that the International Psoriasis Council will address elsewhere (Blauvelt *et al.*, manuscripts in preparation).

Laboratory creation of biosimilars

A biosimilar biological medication is required to have the same amino acid sequence as the originator biologic, which is readily known. Recombinant DNA is first created and placed within a plasmid that encodes the same amino acid sequence as the originator biologic. This plasmid is then transfected into a cell line that will produce the new recombinant protein – the new biosimilar. The recombinant protein is then collected and purified from cell-free supernatants.[5] Each of these steps (Fig. 1) requires optimization and precise controls. For example, the choice of the cell line, the culture media, the culture temperature and the purification processes can all be altered, with each change potentially affecting the quality of the end product.[6]

Analytical methods utilized to define biosimilarity

Biologically produced pharmaceuticals, monoclonal antibodies in many cases, are large, complex proteins (approximately 150 kDa). The primary amino acid composition of a biosimilar medication is precisely bioengineered, but other features of biologics such as three-dimensional protein folding, glycosylation, charge and presence of impurities are more variable during the manufacturing process.[7,8] These particular features of a biologically produced product may affect both the antigen binding and immunogenicity of a given drug, and thus may affect both drug efficacy and safety in clinical use.[9] Approximately 40 different analytical methods are utilized to assess approximately 100 different drug attributes. In total, these analytical studies are considered to be the foundation of approval for biosimilars, or the 'base of the pyramid' (Fig. 2).[10]

Evaluating post-translational modifications via mass spectrometry, including testing for glycosylation, acetylation, sulfation, phosphorylation, glycation and charge, is essential in the characterization of biologics and biosimilars.[11,12] The choice of cell line and cell-culture conditions for drug production greatly affects these parameters. Because of this, biological companies have closely monitored post-translational protein modifications as they have changed manufacturing processes

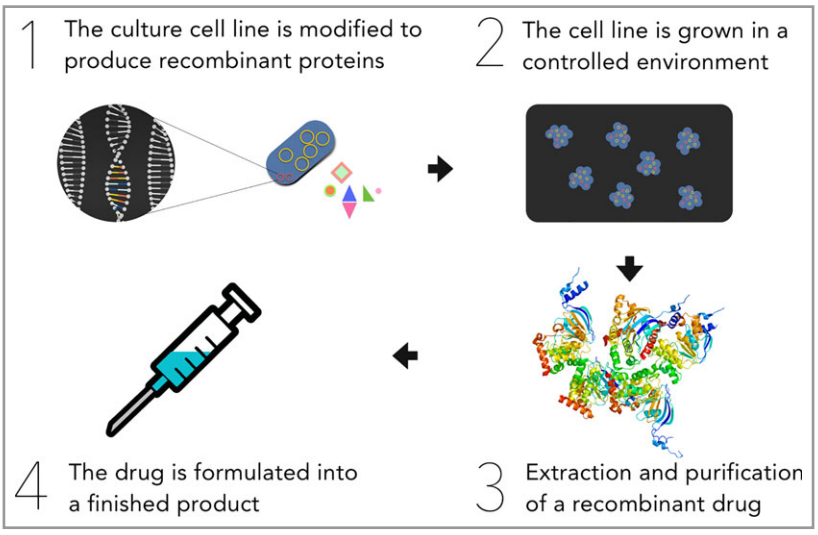

Fig 1. Schematic outlining how biosimilars are created and tested in the laboratory prior to clinical use.

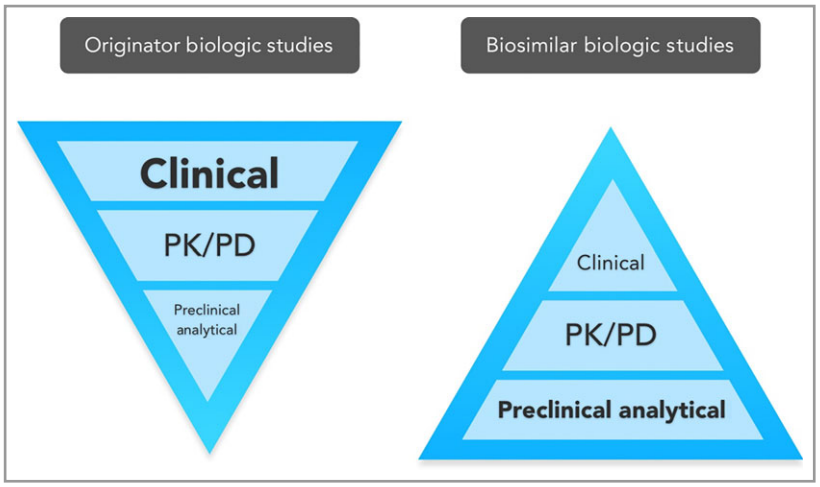

Fig 2. Preclinical assessments form the foundation of the 'pyramid' of biosimilar development. PK/PD, pharmacokinetics/pharmacodynamics.

over time. Drug companies are required to notify the Food and Drug Administration (FDA) regarding each manufacturing change, as well as to provide protein bioanalytical data (e.g. glycosylation status), which characterizes and compares batches of drug before and after each process change.[13] Interestingly, the FDA assumes that relatively minor changes in protein biochemistry will not translate into major changes in drug efficacy and safety for patients, as no new clinical studies have been required when drug manufacturing changes have occurred.

Using cation exchange chromatography and glycan mapping, Schiestl *et al.*[8] showed that the biochemical fingerprint of marketed Enbrel® products produced prior to and after 2009 varied considerably (by 20–40%) in terms of the number of basic variants (e.g. C-terminal lysine variants) and the degree of glycosylation. The changes were attributed to variations in the manufacturing process of Enbrel.[14] In other words, etanercept marketed as Enbrel and produced many years ago is not identical to Enbrel produced today. Thus, it is interesting to posit that the pharmaceutical industry and regulating authorities have already been in the business of making and approving biosimilars (while not naming them in this manner) for many years now.

It is not surprising then that current companies involved in producing biosimilars have argued that detailed analytical studies, which have improved greatly over time, are all that are needed in order to gain approval of their drugs. In other words, if the original companies producing parent biologics are not required to redo clinical studies with each manufacturing change, then the biosimilar companies should also not be required to perform clinical studies to gain initial approval. Although it is true that defining biosimilarity solely on analytical methods would dramatically reduce the costs of developing biosimilar products, most regulatory authorities have continued to rely upon clinical data, albeit limited in some cases, in their approval processes.[5] In one enlightening case, the European Medicines Agency (EMA) rejected a biosimilar interferon alpha 2 product that fulfilled preclinical analytical biosimilarity criteria, but was shown to be different when compared with the originator biological in clinical trials.[15]

Preclinical functional assays are also key when assessing similarity between biosimilars and their originator compounds.[16] Comparisons of drug binding affinity and avidity to the target, for example tumour necrosis factor (TNF)-α, need to be assessed. The ability to neutralize target cytokine activity in cell-culture-based assays is critical. One biological function of TNF blockers, antibody-dependent cell-mediated cytotoxicity (ADCC), is believed to be critical for efficacy in patients with inflammatory bowel disease. Interestingly, Health Canada, the drug-regulating authority in Canada, recently reached a different conclusion from the EMA regarding approval of a biosimilar infliximab product (marketed under the names Remsima® and Inflectra®). Remsima/Inflectra were approved for use in inflammatory bowel diseases by the EMA, but not by Health Canada.[17] Health Canada argued that ADCC had not been adequately demonstrated in the laboratory for the biosimilar infliximab product that underwent review. This case highlights the fact that preclinical characterization of biological function, or lack thereof, can contribute later on to important approval and indication decisions made by regulatory authorities.

Lastly, adequate characterization of the end product to be used in humans is essential for biosimilar companies, as it is for all pharmaceutical companies. This includes analysis of excipients, impurities and aggregates.[18] Again, the choice of cell line, cell-culture parameters and purification processes greatly influence these quality parameters. Testing for drug product stability (e.g. shelf life and alterations with temperature) and product devices (e.g. autoinjectors, prefilled syringes) are also needed in order to determine similarity between the biosimilar product and originator biological product.

Suggestions for preclinical characterization of biosimilars

The EMA, the FDA and Health Canada have stated that they have made and will continue to make approval and indication decisions on biosimilars based on the total evidence of similarity presented by biosimilar companies.[19,20] In other words,

the biosimilar product should not show any meaningful differences from the reference medicine in terms of its quality, safety and efficacy. Biosimilar applications, among other things, must include extensive preclinical analytical data on the structure and function of the drugs. Indeed, the FDA and EMA have issued guidelines for biosimilar companies to follow in order to determine preclinical biosimilarity.[21–23]

Preclinical analysis is the first step within the stepwise approach suggested by the FDA and EMA guidelines and is considered to be the foundation for regulatory approval of biosimilars by both agencies. The FDA and EMA guidelines request investigation of several quality factors including expression system, manufacturing system, physicochemical properties, functional assays, receptor binding, immunochemical properties, finished product stability and impurities. The guidelines recommend that these key quality attributes undergo evaluation with state-of-the-art methods, and in some cases certain attributes should be analysed with multiple methods that are complementary in order to maximize the sponsor's ability to detect and characterize any differences observed between the reference medicine and biosimilar product. In addition, the regulatory agencies encourage the use of orthogonal quantitative methods to characterize differences between the reference product and biosimilar product. Overall, the FDA and EMA guidelines are aligned with regards to the requirements necessary for the demonstration of comparable quality, efficacy and safety between the reference product and biosimilar product. The updated EMA guideline stresses the importance of sensitive *in vitro* comparative assays to evaluate target binding, signal transduction and functional activity of the product.

However, these regulatory statements are somewhat vague in that they do not precisely define the exact types of tests required, the internal quality standards and controls, and the exact degree of variability allowed within individual tests. Acceptance criteria for each quality attribute are not set, and any allowable differences between the reference medicine and biosimilar product are not clearly defined.

Because of this, it is likely that representative internal company policies differ greatly in how they label drugs 'similar' in a given assay. When compared with originator drugs, should biosimilar assay results be within 1 SD, 2 SDs, 80–125% or some other statistical range? In fact, standardization of biosimilar preclinical analyses and their accepted variances are lacking. This might open the door to future approval of biosimilar drugs of poor quality and/or drugs that have not been adequately tested.[24,25]

To provide better assurance of high-quality biosimilars in the future, the International Psoriasis Council suggests that companies involved in producing biosimilars follow a minimum set of analytical standards (a 'biosimilarity index') in the preclinical characterization of their compounds (Table 1). These standards should include the following: (i) confirmation that the amino acid sequence of the biosimilar is identical to that of the originator biologic; (ii) analysis of post-translational modifications, such as glycosylation, acetylation, sulfation, phosphorylation and glycation, which should be repeated regularly over time, especially with changes in manufacturing procedures; (iii) assessment of drug charge; (iv) assessment of the binding affinity of the drug to the target; (v) performance of relevant assays to determine biological function; (vi) analysis of excipients, impurities and aggregates; and (vii) testing for end-product drug stability and delivery device.

Table 1 Proposed International Psoriasis Council Biosimilarity Index: comparisons between biosimilars and the originator biologics

Amino acid sequence
Post-translational modifications, e.g. glycosylation
Drug charge
Drug binding affinity to target
Assays to determine biological function
Analysis of excipients, impurities and aggregates
End-product drug stability and delivery device

The index would be an algorithm where each comparison is weighted with regard to its criticality, and where variability for each assay/test (e.g. < 10% or ± 1 SD) is standardized. Using this index, biosimilars would be rated and scored regarding their preclinical analytical similarity to the originator biologic.

Such an index, which should include a weighting factor for each attribute, as well as accepted variances for each parameter, would provide manufacturing guidance to biosimilar companies; regulatory authorities could enforce compliance with such standards, thus assuring better quality control for emerging biosimilars. This biosimilarity index is comparable in its content to the preclinical list of quality factors for consideration as described by the FDA and EMA; however, the index differs in its execution as it is conceptualized to use predefined acceptance parameters and weighting for analytical tests. The proposed strategy aims to enhance the sponsor's ability to detect and track any potential differences in the preclinical data that may affect downstream decisions regarding *in vivo* and clinical studies. Lastly, it should be emphasized that the International Psoriasis Council's purpose here is to introduce the general concept of such a standardized index, not to provide a definitive tool that is ready for use. Details of its final composition could be discussed and debated among scientists, mathematicians, drug manufacturers and regulatory authorities.

Conclusions

The promise of biosimilars is that these drugs will provide safe, effective and less costly alternatives to patients with psoriasis compared with the originator biologics. It is our hope that decreased costs of biosimilars will lead to increased access of these drugs to needy patients with psoriasis who might otherwise not have been treated with these life-changing drugs. Decreasing the costs of developing biosimilars translates into fewer clinical studies being performed prior to regulatory authority approval. Because fewer

clinical studies will be performed, regulatory authorities and prescribing dermatologists need to rely upon preclinical analytical studies to define similarity between a biosimilar and an originator biologic. The International Psoriasis Council suggests guidelines for standardization of preclinical assessments of emerging biosimilars through the development of a biosimilar index that can be followed by manufacturing companies and enforced by regulatory authorities. Meeting such standards may best assure the creation of safe, effective and high-quality biosimilar drugs for our patients with psoriasis.

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